

Table S4: Primer pairs used to amplify some of the modern

mitogenomes. Ta = annealing temperature, Ext. = extension time, Size = size of the amplicon generated by the primer pair, Location = a rough estimate of the part of the mitogenome the primers amplify based on the horse reference mitogenome (JN398398). * SF and LF are universal mammalian mitogenome amplifying primers (designed in-house by Sandra Abel Nielsen), Pr1 and Pr2 are from study Xu et al. [19], and Pr3 was designed in-house by Ludovic Orlando.

Primer pair	Primer sequence	Ta °C	Ext.	Size (bp)	Location bp
LF_Fwd*	5'-CCGCCTGTTTACCAAAAACATCACC-3'	59	16min	15,100	1,980-580
LF_Rev*	5'-CGAATTTTGARTTTCCTGRACCGCC-3'				
SF_Fwd*	5'-GCTTAAACTYAAAGGACYTGGCGG-3'	59	1min 30s	1,400	580-1,980
SF_Rev*	5'-GGCGGACAAATGGTTTTGTAGTGG-3'				
Pr3_Fwd*	5'-CTCCCAAAGCCCATGTAGA-3'	50	6min 30s	6,500	10,800-750
Pr3_Rev*	5'-GYCTACACCTTGACCTAACG-3'				
LF_Fwd	see above	50	11min	10,900	1,900-12,800
Pr3.2_Rev	5'-TGCTCGTCATTTAGGCTGTG-3'				
6.3_Fwd	5'-CACATCAGCTACCATAATCATCGC-3'	50	5min	5,000	6,300-11,300
11.3_Rev	5'-GGTTAGGCTGGCTAATAGTCATC-3'				
1.6_Fwd	5'-GCAGCCATCAATTAAGAAAGCG-3'	53	6min 30s	6,200	1,600-7,800
7.8_Rev	5'-CATGTTGATGTATCCAAGTGGC-3'				
10.5_Fwd	5'-CCGCCACAGAACTAATCTCC-3'	50	5min	5,000	10,500-15,400
15.4_Rev	5'-GTTGATGGTGGAGCTAGAGC-3'				
14.3_Fwd	5'-CCTAGGAATCTGCCTAATCC-3'	48	3min	3,000	14,300-750
Pr3_Rev	see above				
Pr1_Fwd*	5'-TAACATGAATCGGCGGACA-3'	57	90s	2,000	15,190-700
Pr1_Rev*	5'-TTGCTGAAGATGGCGGTAT-3'				
Pr2_Fwd*	5'-ATTTCCATAGACAGGCATCC-3'	57	90s	1,500	16,600-1,440
Pr2_Rev*	5'-TCACCTCTACCTACGAATCTTCT-3'				
MAC_16.2_Fwd	5'-ATTACTCCGCATCAGCAACC-3'	53	30s	200	16,200-16,400
MAC_16.4_Rev	5'-GGGGGTTTGATTARGAAARTTACA-3'				
MAC_1.4_Fwd	5'-TGTCGCAAAATAGTGAGAAGATTT-3'	53	30s	200	1,400-1,600
MAC_1.6_Rev	5'-TGGCTGCTTTTAAGCCAAC-3'				
MAC_16.5_Fwd	5'-ACCCCRAAAACAAGACYAAA-3'	53	30s	100	16,500-16,600
MAC_16.6_Rev	5'-TGYAGGTTTGGAAAATCAGA-3'				
MAC_1.8_Fwd	5'-CAACCCAAAACAACTAACCACT-3'	53	30s	200	1,800-2,000
MAC_2.0_Rev	5'-TTTGCACGGTTAGGATACCG-3'				
0.7rev	5'-GATTAAGTCGGATATATGGCGG-3'	(These were designed for sequencing at MacroGen together with the SF.Fwd and SF.Rev primers, and were never amplified in our laboratory)			
1.0fwd	5'-GTATACCGGAAGGTGTAAGTGG-3'				
1.4rev	5'-CTTGTTGAGTAGATACAGCG-3'				
1.8fwd	5'-GTTAACCCAACACAGGCATG-3'				